

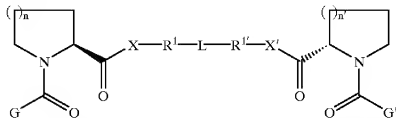
# AMENDMENTS TO THE CLAIMS

This listing of the claims replaces all previous claim listings in the application.

1. (Currently Amended) A method for rendering a subpopulation of mammalian hematopoietic stem cells susceptible to divalent ligand-induced growth, proliferation or differentiation, which method comprises

transducing one or more cells of a population of mammalian primary hematopoietic stem cells with at least one retroviral vector comprising at least one recombinant DNA construct encoding a fusion protein which comprises at least one signaling domain derived from an intracellular portion of a thrombopoietin receptor and at least one ligand-binding domain derived from F36V which is heterologous with respect to the signaling domain and binds to a selected divalent ligand capable of inducing association of two or more molecules of F36V

such that upon exposure of the transduced cells to a concentration of the divalent ligand having the formula:

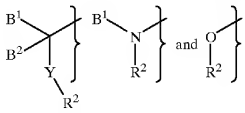


effective to induce association of two or more of the encoded fusion proteins, growth, proliferation or differentiation of said cells is induced;

wherein X and X' can be O, NH, or CH<sub>2</sub>;

L is a covalently linker moiety;

wherein G and G' are independently selected from the group comprised of



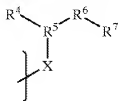
B<sup>1</sup> and B<sup>2</sup> are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkynyl, substituted aryl, aryl, or heteroaryl moieties;

Y is O, S, NH, -NH(C=O)-, NH(C=O)-O-, NH(SO<sub>2</sub>)-, NR<sub>3</sub>, or a covalent bond;

R<sup>1</sup>, R<sup>1'</sup>, and R<sup>2</sup> are the same or different and are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkyl, substituted aryl, aryl, or heteroaryl moieties;

n and n' are each independently 1 or 2;

wherein at least one of X-R<sup>1</sup> and X'-R<sup>1'</sup> is independently a moiety:



wherein R<sup>4</sup> is hydrogen; branched, unbranched, cyclic, saturated or unsaturated, substituted or unsubstituted aliphatic; branched, unbranched or cyclic heteroaliphatic; aryl or heteroaryl;

R<sup>5</sup> is a branched, unbranched or cyclic, aliphatic moiety of 1 to 8 carbon atoms;

R<sup>6</sup> is a substituted or unsubstituted aliphatic, heteroaliphatic, heterocyclic, aryl or heteroaryl ~~moiety~~ moiety;

R<sup>7</sup> is hydrogen or a reactive functional group permitting covalent attachment to a linker moiety; and

wherein the transduction is carried out *in vivo* or *ex vivo* and wherein said transduced cells are suitable for introduction into a mammal.

Claims 2-3 (Canceled).

4. (Previously presented) The method of Claim 1, wherein the subpopulation of mammalian primary hematopoietic stem cells comprises at least one of bone marrow cells, cord blood cells, and peripheral blood cell.

5. (Previously presented) The method of Claim 1, wherein the mammalian primary hematopoietic stem cells are human cells.

Claims 6-11 (Canceled).

12. (Previously presented) The method of Claim 1 wherein the cells are removed from the mammal prior to being transduced with the retroviral vector comprising at least one recombinant DNA construct.

13. (Original) The method of Claim 12 which further comprises introducing the transduced cells so obtained into a mammal.

14. (Previously presented) The method of Claim 13 wherein the transduced cells are treated with divalent ligand prior to their introduction into the mammal.

15. (Original) The method of Claim 13 wherein the cells are allogeneic with respect to the mammal.

16. (Original) The method of Claim 13 wherein the cells are syngeneic with respect to the mammal.

17. (Original) The method of Claim 13 wherein the cells are autologous with respect to the mammal.

18. (Original) The method of Claim 13 wherein the mammal is a human.

19. (Original) The method of Claim 1 wherein the cells are transduced within the mammal.

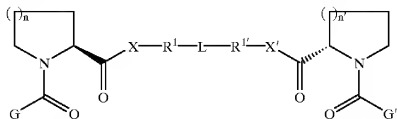
Claim 20 (Canceled).

21. (Currently Amended) A method for expanding a subpopulation of mammalian hematopoietic stem cells comprising:

(a) providing a subpopulation of mammalian primary hematopoietic stem cells which has been transduced with at least one retroviral vector comprising at least one recombinant DNA construct encoding a fusion protein which (i) comprises at least one signaling domain derived from a thrombopoietin receptor and at least one divalent ligand-binding domain derived from F36V, and (ii) induces growth, proliferation or differentiation upon multimerization with one or more other fusion proteins containing at least one signaling domain; and

(b) treating the subpopulation of cells with a concentration of a divalent ligand capable of

inducing association of two or more molecules of F36V having the formula:

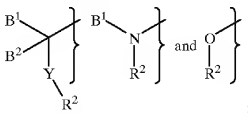


effective to induce association of two or more of the encoded fusion proteins, growth, proliferation or differentiation of said cells is induced;

wherein X and X' can be O, NH, or CH<sub>2</sub>;

L is a covalently linker moiety;

wherein G and G' are independently selected from the group comprised of



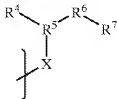
B<sup>1</sup> and B<sup>2</sup> are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkynyl, substituted aryl, aryl, or heteroaryl moieties;

Y is O, S, NH, -NH(C=O)-, NH(C=O)-O-, NH(SO<sub>2</sub>)-, NR<sub>3</sub>, or a covalent bond;

R<sup>1</sup>, R<sup>1'</sup>, and R<sup>2</sup> are the same or different and are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkyl, substituted aryl, aryl, or heteroaryl moieties;

n and n' are each independently 1 or 2;

wherein at least one of X-R<sup>1</sup> and X'-R<sup>1'</sup> is independently a moiety:



wherein R<sup>4</sup> is hydrogen; branched, unbranched, cyclic, saturated or unsaturated, substituted or unsubstituted aliphatic; branched, unbranched or cyclic heteroaliphatic; aryl or heteroaryl;

R<sup>5</sup> is a branched, unbranched or cyclic, aliphatic moiety of 1 to 8 carbon atoms;

R<sup>6</sup> is a substituted or unsubstituted aliphatic, heteroaliphatic, heterocyclic, aryl or heteroaryl moiety;

R<sup>7</sup> is hydrogen or a reactive functional group permitting covalent attachment to a linker moiety; and

wherein said treatment with said divalent ligand is carried out *in vivo* or *ex vivo*, and wherein said transduced cells are suitable for introduction into a mammal.

Claims 22-23 (Canceled).

24. (Previously presented) The method of Claim 21, wherein the subpopulation of mammalian primary hematopoietic stem cells comprises at least one of bone marrow cells, cord blood cells, and peripheral blood cells.

25. (Previously presented) The method of Claim 21, wherein the mammalian primary hematopoietic stem cells are human cells.

Claims 26-31 (Canceled).

32. (Previously presented) The method of Claim 21 wherein the subpopulation of mammalian primary hematopoietic stem cells which has been transduced with the at least one retroviral vector was transduced *ex vivo*.

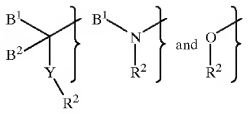
33. (Original) The method of Claim 32 which further comprises introducing the transduced cells so obtained into a recipient mammal.

34. (Previously presented) The method of Claim 33 wherein the transduced cells are treated with the divalent ligand prior to their introduction into the recipient mammal.

35. (Original) The method of Claim 33 wherein the cells are allogeneic with respect to the mammal.

36. (Original) The method of Claim 33 wherein the cells are syngeneic with respect to the mammal.





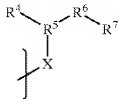
B<sup>1</sup> and B<sup>2</sup> are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkynyl, substituted aryl, aryl, or heteroaryl moieties;

Y is O, S, NH, -NH(C=O)-, NH(C=O)-O-, NH(SO<sub>2</sub>)-, NR<sub>2</sub>, or a covalent bond;

R<sup>1</sup>, R<sup>1'</sup>, and R<sup>2</sup> are the same or different and are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkyl, substituted aryl, aryl, or heteroaryl moieties;

n and n' are each independently 1 or 2;

wherein at least one of X-R<sup>1</sup> and X'-R<sup>1'</sup> is independently a moiety:



wherein R<sup>4</sup> is hydrogen; branched, unbranched, cyclic, saturated or unsaturated, substituted or unsubstituted aliphatic; branched, unbranched or cyclic heteroaliphatic; aryl or heteroaryl;

R<sup>5</sup> is a branched, unbranched or cyclic, aliphatic moiety of 1 to 8 carbon atoms;

R<sup>6</sup> is a substituted or unsubstituted aliphatic, heteroaliphatic, heterocyclic, aryl or heteroaryl ~~moiety moiety~~; and

R<sup>7</sup> is hydrogen or a reactive functional group permitting covalent attachment to a linker moiety.

Claim 58 (Canceled).

59. (Original) A method for treating or preventing a hemopoietic disease or pathological condition in a mammal, comprising expanding a subpopulation of hemopoietic cells by the method of Claim 24 and introducing the resultant cells to the mammal.

Claims 60 - 88 (Canceled).

89. (Previously presented) The method according to claim 1, wherein said divalent ligand is selected from the group consisting of AP1903, AP20187, and AP1510.

90. (Previously presented) The method of claim 21, wherein said divalent ligand is selected from the group consisting of AP1903, AP20187, and AP1510.

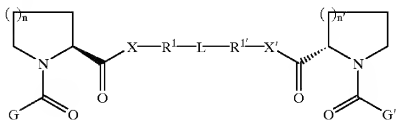
91. (Previously presented) The method of claim 57, wherein said divalent ligand is selected from the group consisting of AP1903, AP20187, and AP1510.

92 [[63]]. (Currently amended) A method for expanding an erythroid cell population comprising:

(a) providing CD34+ primary hematopoietic stem cells transduced with a retroviral vector comprising at least one recombinant DNA construct that encodes a fusion protein that

(i) comprises at least one signaling domain derived from a thrombopoietin receptor and at least one divalent ligand-binding domain derived from F36V, and (ii) induces growth, proliferation or differentiation upon multimerization with one or more other fusion proteins containing at least one signaling domain; and

(b) treating the transduced cells with a concentration of a divalent ligand capable of inducing association of two or more molecules of F36V having the formula:

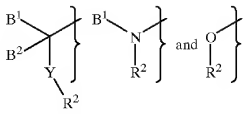


wherein X and X' can be O, NH, or CH<sub>2</sub>;

L is a covalently linker moiety:

wherein G and G' are independently selected from the group comprised of





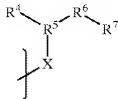
$\text{B}^1$  and  $\text{B}^2$  are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkynyl, substituted aryl, aryl, or heteroaryl moieties;

$\text{Y}$  is O, S, NH,  $\text{-NH(C=O)-}$ ,  $\text{NH(C=O)-O-}$ ,  $\text{NH(SO}_2\text{)-}$ ,  $\text{NR}_3$ , or a covalent bond;

$\text{R}^1$ ,  $\text{R}^{1'}$ , and  $\text{R}^2$  are the same or different and are each independently hydrogen, alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heteroalkyl, heteroalkenyl, heteroalkynyl, heterocycloalkyl, heterocycloalkenyl, heterocycloalkyl, substituted aryl, aryl, or heteroaryl moieties;

$n$  and  $n'$  are each independently 1 or 2;

wherein at least one of  $\text{X-R}^1$  and  $\text{X'-R}^{1'}$  is independently a moiety:



wherein  $\text{R}^4$  is hydrogen; branched, unbranched, cyclic, saturated or unsaturated, substituted or unsubstituted aliphatic; branched, unbranched or cyclic heteroaliphatic; aryl or heteroaryl;

$\text{R}^5$  is a branched, unbranched or cyclic, aliphatic moiety of 1 to 8 carbon atoms;

$\text{R}^6$  is a substituted or unsubstituted aliphatic, heteroaliphatic, heterocyclic, aryl or heteroaryl moiety; and

$\text{R}^7$  is hydrogen or a reactive functional group permitting covalent attachment to a linker moiety;

wherein said treatment with said divalent ligand is carried out *in vivo* or *ex vivo*;

wherein said treatment results in expansion of erythroid cells from the transduced cells; and

wherein said transduced cells are suitable for introduction into a mammal.

93 ~~[[64]]~~. (Currently amended) The method of claim 92 ~~[[63]]~~, wherein said divalent ligand is selected from the group consisting of AP1903, AP20187, and AP1510.